

Final report-Teratologic Evaluation of FDA 71-23 (Amaranth: FD&C Red #2) in Mice,  
Rats, Hamsters & Rabbits  
2/15/72

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**FOOD AND DRUG  
Research LABORATORIES, INC.**

MAURICE AVENUE AT 58TH STREET, MASPETH, NEW YORK 11378



February 15, 1972

**Teratologic Evaluation of FDA 71-23**

**(Amaranth; FD & C Red No. 2)**

**in**

**Mice, Rats, Hamsters and Rabbits**

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## FINAL REPORT

Submitted to: DHEW/Public Health Service  
Food and Drug Administration CA-272  
5600 Fishers Lane-Room 5C-13  
Rockville, Maryland 20852

Date: February 15, 1972

Laboratory No. 0728 g  
Contract No. FDA 71-260

Sample: Fine dark purple material.

Marking: FDA 71-23 (Amaranth; FD & C Red No. 2).

Examination Requested: Teratologic evaluation of FDA 71-23 in mice.

:

Procedure: See Appendix I

Results: See Tables 1 through 4 and Appendix II

Conclusion: Attention is called to the fact that this is the first of a series of reports which will be issued in accordance with the terms of the contract cited above. Eventually, a total of at least 36 compounds will have been tested in 18 pairs; each pair being run concurrently against one sham-treated control and one positive control group. Because of the inherent variability of biological data of the type dealt with here, the accumulation and pooling of sequential sets of control values will greatly enhance the statistical value of the findings and the ultimate reliability of the test results.

For these reasons, the conclusion stated below is regarded as provisional and subject to reexamination in the light of later findings:

"The administration of up to 1000 mg/kg (body weight) of the test material to pregnant mice for 10 consecutive days had no clearly discernible effect on nidation or on maternal or fetal survival. The number of abnormalities seen in either soft or skeletal tissues of the test groups did not differ from the number occurring spontaneously in the sham-treated controls."

FOOD AND DRUG RESEARCH LABORATORIES, INC.

*Kenneth Morgareidge*  
Kenneth Morgareidge, Ph.D.  
Vice President

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## FOOD AND DRUG RESEARCH LABORATORIES, INC.

Groups: 1 &amp; 2; 7 through 10

Material: FDA 71-23

Table 1

Fate Summary  
( Mice )Date February 15, 1972  
Laboratory No. 0728 g

Group	Material	Dose mg/kg	Mated	Total	Pregnant	Surviving (Total) Number Pregnant	
						At Term	Number Pregnant
1	Sham	0	27	20	25	19	
2	Aspirin*	200	30	26	20	14	
7	FDA 71-23	27	24	21	23	20	
8	FDA 71-23	90	25	21	24	20	
9	FDA 71-23	300	23	20	23	20	
10	FDA 71-23	1000	26	21	25	21	

\* Positive Control

Group: 1 & 2; 7 through 10

Material: FDA 71-23

FOOD AND DRUG RESEARCH LABORATORIES, INC.

Date February 15, 1972

Table 2

Reproduction Data  
( Mice )

Group:	1	2	Aspirin*	7	8	9	10
Dose (mg/kg):	Sham			27	90	300	1000
Number of females:							
Total pregnant	20	26		21	21	20	21
Pregnant at term	19	14		20	20	20	21
Number of live litters:	20	14		20	20	20	21
Number of implant sites:							
Total	247	308		283	270	249	274
Average/pregnant dam	12.4	11.8		13.5	12.9	12.5	13.0
Number of fetuses alive:							
Total	240	165		254	248	239	258
Average/live litter	12.0	11.8		12.7	12.4	12.0	12.3
Average/pregnancy at term	12.6	11.8		12.7	12.4	12.0	12.3
Number of fetuses dead:							
Total	5	49		19	5	1	2
Litters with one or more dead	4	6		5	4	1	2
Pregnancies at term (%)	21.1	42.9		25.0	20.0	5.0	9.5
Litters with all dead	0	4		0	0	0	0
Pregnancies at term (%)	0	28.6		0	0	0	0
Number of resorptions:							
Total:	2	94		10	17	9	14
Litters with one or more resorptions	1	12		7	7	8	9
Pregnancies at term (%)	5.3	85.7		35.0	35.0	40.0	42.9
Litters with total resorptions	0	8		0	1	0	0
Pregnancies at term (%)	0	57.1		0	5.0	0	0
Average fetus weight, g.	0.97	0.87		0.94	0.99	0.94	1.00

\* Positive control at 200 mg/kg

Groups 1&2; 7 through 10  
 Material FDA 71-23

FOOD and DRUG RESEARCH LABORATORIES, INC.

Table 3  
 Summary of Skeletal Findings

Laboratory No. 0728 8  
 Date February 15, 1972

Findings	Group No:	1	2	7	8	9	10
	Dose (mg/kg):Sham		Aspirin	27	90	300	1000
<u>Live Fetuses Examined</u>		163/19	115/14	181/20	179/20	178/20	182/21
<u>Fetuses with:</u>							
Incomplete sternebrae	14/6	20/7	27/10	56/13	45/13	50/15	
Scrambled sternebrae	123/17	75/14	112/18	63/17	95/18	88/15	
Bipartite sternebrae							1/1
Missing sternebrae							
Fused sternebrae	3/3	1/1	1/1	1/1	1/1	1/1	
Extra sternebrae			6/2	1/1	12/5	12/5	
Incomplete hyoid							
<u>Fetuses with:</u>							
Scrambled vertebrae							
Tail defects (short, scrambled, etc.)	137/17	57/10	113/17	82/17	106/17	34/11	
Incomplete ossification; vertebrae							1/1
Spinal bifida							
<u>Fetuses with:</u>							
Fused ribs							
Incomplete ribs							1/1
Wavy ribs							
Less than 12 ribs	1/1						
More than 13 ribs	2/2	5/2	18/9	1/1	5/2	10/5	
<u>Other Findings:</u>							
Scoliosis	25/7	32/9	37/11	41/11	91/15	19/6	
Delayed cranial ossification							
Craniosostosis	115/16	35/4	118/15	126/14	159/20	164/20	
Feet; retarded ossification							
Underdeveloped/missing occipital							21/4

\* Numerator = Number of fetuses affected; Denominator = number of litters affected

## FOOD AND DRUG RESEARCH LABORATORIES, INC.

Groups 1 &amp; 2; 7 through 10

Species Mice

Date February 15, 1972

Laboratory No. 0728 q

Table 4

Average Body Weights \*

Group	Material	Dose Level	0	6	11	Day 15	Day 17**
		mg/kg	g				
1	Sham	0	29.3	31.4	34.6	44.1	49.6 (19)
2	Aspirin	200	28.8	31.0	31.1	36.1	44.8 (14)
7	FDA 71-23	27	29.7	31.7	36.1	44.8	51.8 (20)
8	FDA 71-23	90	29.6	32.7	36.2	45.2	51.6 (20)
9	FDA 71-23	300	29.9	32.5	35.7	43.5	48.2 (20)
10	FDA 71-23	1000	30.1	32.4	36.2	45.6	52.0 (21)

\* Of pregnant dams.

\*\* Number of surviving dams in parentheses (c.f. Table 1).



## Appendix I

### Teratology Study in Mice

Virgin adult female albino CD-1 outbred mice were individually housed in disposable plastic cages in temperature and humidity-controlled quarters with free access to food and fresh tap water. They were mated with young adult males, and observation of the vaginal sperm plug was considered Day 0 of gestation. Beginning on Day 6 and continuing daily through Day 15 of gestation, the females were dosed with the indicated dosages by oral intubation; the controls were sham treated.

Body weights were recorded on Days 0, 6, 11, 15, and 17 of gestation. All animals were observed daily for appearance and behavior with particular attention to food consumption and weight, in order to rule out any abnormalities which may have occurred as a result of anorexic effects in the pregnant female animal.

On Day 17 all dams were subjected to Caesarean section under surgical anesthesia, and the numbers of implantation sites, resorption sites, and live and dead fetuses were recorded. The body weights of the live pups were also recorded. The urogenital tract of each dam was examined in detail for anatomical normality.

All fetuses were examined grossly for the presence of external congenital abnormalities. One-third of the fetuses of each litter underwent detailed visceral examinations employing 10X magnification. The remaining two-thirds were cleared in potassium hydroxide (KOH), stained with alizarin red S dye and examined for skeletal defects.

## FOOD AND DRUG RESEARCH LABORATORIES, INC.

Group 1  
Material Sham  
Dose 0

## Appendix II

## Reproduction Data in Mice (Individual)

Date February 15, 1972  
Laboratory No. 0728

Dam No.	Fate *	Implant Sites	Alive	Dead	Fetuses	Resorption Sites	Average Fetus Weight (g)	Remarks
S 8001	P	18	18	0	0	0	0.91	
S 8002	P	12	12	0	0	0	0.89	
S 8003	P	15	15	0	0	0	0.63	
S 8004	NP	0					--	Died Day 16
S 8005	P	14	12	2	0	0	1.05	
S 8006	P	14	14	0	0	0	0.96	
S 8007	P	14	14	0	0	0	1.03	
S 8008	P	9	9	0	0	0	0.79	
S 8009	P	11	11	0	0	0	1.31	
S 8010	P	12	11	1	0	0	0.95	
S 8011	P	13	13	0	0	0	0.89	
S 8012	NP	0					--	
S 8013	P	10	10	0	0	0	0.87	
S 8014	NP	0					--	
S 8015	P	13	13	0	0	0	0.85	
S 8016	P	15	15	0	0	0	0.75	
S 8017	P	11	11	0	0	0	1.00	
S 8018	P	12	11	1	0	0	0.81	
S 8019	P	9	9	0	2	2	--	Dam died day 14
S 8020	P	16	14	0			0.98	
S 8021	NP	0					--	
S 8022	NP	0					--	
S 8023	NP	0					--	
S 8024	NP	0					--	
S 8025	P	6	6	0	0	0	1.53	
S 8026	P	13	13	0	0	0	1.15	
S 8027	P	10	9	1	0	0	1.12	

\* P= Pregnant; NP= Not Pregnant

## FOOD AND DRUG RESEARCH LABORATORIES, INC.

Group 2Material AspirinDose 200 mg/kgDate February 15, 1972  
Reproduction Data in Mice (Individual) Laboratory No. 0728

## Appendix II

Dam No.	Fate *	Implant Sites	Fetuses		Resorption Sites	Average Fetus Weight (g)	Remarks
			Alive	Dead			
A 8001	P	12	0	12	0	0.74	Died day 15.
A 8002	P	10	0	0	0	--	
A 8003	NP	0	0	12	7	--	Died day 16.
A 8004	P	12	0	0	2	--	
A 8005	P	7	0	0	1	0.80	
A 8006	P	2	0	0	13	--	
A 8007	P	14	0	0	0	0.69	
A 8008	P	13	0	0	13	--	
A 8009	P	11	0	0	11	--	Died day 12.
A 8010	P	16	0	0	16	--	Died day 17.
A 8011	P	13	10	0	3	0.69	Died day 12.
A 8012	P	13	0	0	13	--	Died day 11.
A 8013	P	14	0	0	14	--	Died day 14.
A 8014	P	13	13	0	0	0.74	
A 8015	P	12	0	12	0	--	
A 8016	P	12	11	0	1	0.92	
A 8017	P	15	0	15	0	--	
A 8018	P	12	12	0	0	1.11	
A 8019	P	8	0	8	0	--	
A 8020	NP	0	0	0	0	0.81	
A 8021	P	12	11	1	0	--	
A 8022	NP	0	0	0	0	0.83	
A 8023	P	14	14	0	0	1.08	
A 8024	P	11	11	0	0	0.82	
A 8025	P	8	8	0	0	0.88	
A 8026	P	14	14	0	0	0.89	
A 8027	NP	0	0	0	0	1.03	
A 8028	P	15	14	1	0	0.89	
A 8029	P	12	12	0	0	1.03	
A 8030	P	13	12	0	1	0.87	

\* P= Pregnant; NP= Not Pregnant

## FOOD AND DRUG RESEARCH LABORATORIES, INC.

Group 7Material FDA 71-23Dose 27 mg/kgDate February 15, 1972  
Reproduction Data in Mice (Individual)      Laboratory No. 0728 g

## Appendix II

## Remarks

Dam No.      Fate \*      Implant Sites      Fetuses      Resorption Sites      Average Fetus Weight (g)

Alive      Dead

G 8001	P	13	0	12	1	--
G 8002	P	11	10	1	1	0.78
G 8003	P	14	14			0.91
G 8004	P	11	10	1	1	1.25
G 8005	P	18	12	4	2	0.80
G 8006	NP	0				--
G 8007	P	14	12	1	1	
G 8008	P	16	15	1		
G 8009	P	14	14			0.88
G 8010	P	14	14			0.88
G 8011	P	9	9			0.88
G 8012	P	17	17			0.98
G 8013	P	12	12			0.89
G 8014	P	12	9			1.12
G 8015	P	15	14	1	1	0.79
G 8016	P	16	16			0.86
G 8017	NP	0				--
G 8018	P	10	10			--
G 8019	P	13	13			1.01
G 8020	P	13	12			1.09
G 8021	NP	0			1	0.98
G 8022	P	13	13			--
G 8023	P	13	13			0.88
G 8024	P	15	15			1.03
						0.95

Died Day 14.

\* P= Pregnant; NP= Not Pregnant

## FOOD AND DRUG RESEARCH LABORATORIES, INC.

Group 8                                  Date February 15, 1972  
 Material FDA 71-23                          Laboratory No. 0728 g  
 Dose 90 mg/kg

## Appendix II

## Reproduction Data in Mice (Individual)

Dam No.	Fate *	Implant Sites	Alive	Dead	Fetuses	Resorption Sites	Average Fetus Weight (g)	Remarks
G 8031	P	16	15				0.96	
G 8032	NP	0					--	
G 8033	P	14	14				0.84	
G 8034	P	11	11				0.99	
G 8035	NP	0					--	
G 8036	P	15	15				1.24	
G 8037	NP	0					--	
G 8038	P	11	11				0.98	
G 8039	P	11	9	2			0.96	
G 8040	P	13	13				1.03	
G 8041	P	14	13	1			0.85	
G 8042	P	15	12	1	2		1.00	
G 8043	P	13	12	1			1.04	
G 8044	P	14	13	1			0.87	
G 8045	P	14	14	1			0.92	
G 8046	P	11	10				0.86	
G 8047	P	14	14				1.08	
G 8048	P	12	11	1			0.99	
G 8049	NP	0					--	
G 8050	P	11	10		1		0.86	
G 8051	P	10				10	--	
G 8052	P	12	12				1.26	
G 8053	P	11	11				1.03	
G 8054	P	11	11				0.94	
G 8055	P	17	17				1.05	

\* P= Pregnant; NP= Not Pregnant

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## FINAL REPORT

Submitted to: DHEW/Public Health Service  
Food and Drug Administration CA-272  
5600 Fishers Lane-Room 5C-13  
Rockville, Maryland 20852

Date: February 15, 1972

Laboratory No. 0729 g  
Contract No. FDA 71-260

Sample: Fine dark purple material.

Marking: FDA 71-23 (Amaranth; FD & C Red No. 2).

Examination Requested: Teratologic evaluation of FDA 71-23 in rats

Procedure: See Appendix I

Results: See Tables 1 through 4 and Appendix II

Conclusion: Attention is called to the fact that this is the first of a series of reports which will be issued in accordance with the terms of the contract cited above. Eventually, a total of at least 36 compounds will have been tested in 18 pairs; each pair being run concurrently against one sham-treated control and one positive control group. Because of the inherent variability of biological data of the type dealt with here, the accumulation and pooling of sequential sets of control values will greatly enhance the statistical value of the findings and the ultimate reliability of the test results.

For these reasons, the conclusion stated below is regarded as provisional and subject to reexamination in the light of later findings:

"The administration of up to 1000 mg/kg (body weight) of the test material to pregnant rats for 10 consecutive days had no clearly discernible effect on nidation or on maternal or fetal survival. The number of abnormalities seen in either soft or skeletal tissues of the test groups did not differ from the number occurring spontaneously in the sham-treated controls."

FOOD AND DRUG RESEARCH LABORATORIES, INC.

Kenneth Morgareidge, Ph.D.  
Vice President

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## FOOD AND DRUG RESEARCH LABORATORIES, INC.

Groups: 1 & 2; 7 through 10Material: FDA 71-23Date February 15, 1972Laboratory No. 0729 g

Table 1

Fate Summary  
( Rats )

Group	Material	Dose mg/kg	Mated	Total	At Term	
					Pregnant	Surviving (Total)
1	Sham	0	24	21	21	25
2	Aspirin*	250	26	20	22	19
7	FDA 71-23	27	24	24	24	24
8	FDA 71-23	90	26	21	21	26
9	FDA 71-23	300	25	22	22	25
10	FDA 71-23	1000	25	22	22	21

\* Positive Control

Group: 1 & 2; 7 through 10  
 Material: FDA 71-23

FOOD AND DRUG RESEARCH LABORATORIES, INC.

Date February 15, 1972

Laboratory No. 0729 q

Table 2

Reproduction Data  
 ( Rats )

	Group:	1	2	Aspirin *	7	8	9	10
	Dose (mg/kg):	Sham			27	90	300	1000
Number of females:								
Total pregnant		21	20		24	21	22	22
Pregnant at term		21	19		24	21	22	21
Number of live litters:		21	19		24	21	22	22
Number of implant sites:								
Total		245	230		254	237	254	227
Average/pregnant dam		11.7	11.5		10.6	11.3	11.5	10.3
Number of fetuses alive:								
Total		242	206		237	225	228	213
Average/live litter		11.5	10.8		9.88	10.7	10.4	9.68
Average/pregnancy at term		11.5	10.8		9.88	10.7	10.4	10.1
Number of fetuses dead:								
Total		0	0		1	0	3	1
Litters with one or more dead		0	1		1	0	2	1
Pregnancies at term (%)		0	5.26		4.17	0	9.09	4.76
Litters with all dead		0	0		0	0	0	0
Pregnancies at term (%)		0	0		0	0	0	0
Number of resorptions:								
Total:		3	15		12	15	23	13
Litters with one or more resorptions		3	8		7	9	10	9
Pregnancies at term (%)		14.3	42.1		37.5	13.3	45.5	42.9
Litters with total resorptions		0	0		0	0	0	0
Pregnancies at term (%)		0	0		0	0	0	0
Average fetus weight, g.		4.05	3.55		4.00	3.84	3.77	3.83

\* Positive control at 250 mg/kg

Groups 1 & 2, 7 through 10  
 Material FDA 71-23

FOOD and DRUG RESE. )CH LABORATORIES, INC.

Groups 1 & 2, 7 through 10

Table 3

Summary of Skeletal Findings  
 (Rats)

Laboratory No. 0729 9  
 Date February 15, 1972

Findings	Group No:	1	2	7	8	9	1000
	Dose (mg/kg):	Sham	Aspirin**	27	90	300	
<u>Live Fetuses Examined</u>		173/21	148/19	176/24	163/21	170/22	147/21
<u>Fetuses with:</u>							
Incomplete sternebrae		101/19	106/19	106/24	102/21	114/21	106/20
Scrambled sternebrae		7/6	2/1	1/1	1/1	1/1	1/1
Bipartite sternebrae		39/12	5/5	6/5	2/2	2/2	10/8
Missing sternebrae			58/14	35/13	39/14	44/15	24/13
Fused sternebrae				1/1			9/3
Extra sternebrae							

Fetuses with:  
Scrambled vertebrae  
 Tail defects (short, scrambled, etc.)

<u>Fetuses with:</u>				
Fused ribs				
Incomplete ribs				
Wavy ribs				
Less than 12 ribs				

<u>Fetuses with:</u>				
Scoliosis				
Delayed cranial ossification				
Craniostosis				
Incomplete ossification of phalanges				

Other Findings:  
 Scoliosis  
 Delayed cranial ossification  
 Craniostosis  
 Incomplete ossification of phalanges

<u>Other Findings:</u>				
Scoliosis				
Delayed cranial ossification				
Craniostosis				
Incomplete ossification of phalanges				

\* Numerator = Number of fetuses affected; Denominator = number of litters affected

## FOOD AND DRUG RESEARCH LABORATORIES, INC.

Groups 1 & 2; 7 through 10Species RatsTable 4 \*  
Average Body Weights  
Species No. 0729 qDate February 15, 1972  
Laboratory No. 0729 q

Group	Material	Dose Level	Day-----g	Day-----g	Day-----g	Day-----g	
		mg/kg	0	6	11	15	20 **
1	Sham	0	240.5	253.0	265.8	281.3	360.2 (21)
2	Aspirin	250	240.1	251.6	258.8	280.1	335.5 (19)
7	FDA 71-23	27	239.3	248.8	263.4	281.8	346.3 (24)
8	FDA 71-23	90	245.6	253.0	266.6	282.6	360.5 (21)
9	FDA 71-23	300	239.5	252.2	262.9	283.6	348.1 (22)
10	FDA 71-23	1000	235.8	250.2	255.0	272.0	328.5 (21)

\* Of pregnant dams

\*\* Number of surviving dams in parentheses (c.f. Table 1).



## Appendix I

### Teratology Study in Rats

Virgin adult female albino rats (Wistar derived stock) were individually housed in mesh bottom cages in temperature and humidity-controlled quarters with free access to food and fresh tap water. They were mated with young adult males, and observation of the vaginal sperm plug was considered Day 0 of gestation. Beginning on Day 6 and continuing daily through Day 15 of gestation, the females were dosed with the indicated dosages by oral intubation; the controls were sham treated.

Body weights were recorded on Days 0, 6, 11, 15, and 20 of gestation. All animals were observed daily for appearance and behavior with particular attention to food consumption and weight, in order to rule out any abnormalities which may have occurred as a result of anorexic effects in the pregnant female animal.

On Day 20 all dams were subjected to Caesarean section under surgical anesthesia, and the numbers of implantation sites, resorption sites, and live and dead fetuses were recorded. The body weights of the live pups were also recorded. The urogenital tract of each dam was examined in detail for anatomical normality.

All fetuses were examined grossly for the presence of external congenital abnormalities. One-third of the fetuses of each litter underwent detailed visceral examinations employing 10X magnification. The remaining two-thirds were cleared in potassium hydroxide (KOH), stained with alizarin red S dye and examined for skeletal defects.

## FOOD AND DRUG RESEARCH LABORATORIES, INC.

Date February 15, 1972

Material FDA 71-1  
Dose Sham

## Reproduction Data in Rats (Individual)

## Appendix II

Laboratory No. 0729

Group	1	Dam No.	Fate *	Implant Sites	Fetuses Alive	Fetuses Dead	Resorption Sites	Average Fetus Weight (g)	Remarks
		S 9001	P	10	9		1	3.81	
		S 9002	P	10	10			4.11	
		S 9003	NP	0				--	
		S 9004	P	11	11			5.86	
		S 9005	P	10	10			3.65	
		S 9006	P	12	12			4.17	
		S 9007	P	16	16			4.16	
		S 9008	P	11	11			3.87	
		S 9009	P	12	12			4.93	
		S 9010	P	12	12			3.56	
		S 9011	P	14	14			3.87	
		S 9012	P	15	15			3.88	
		S 9013	P	12	12			3.71	
		S 9014	P	10	10			3.37	
		S 9015	P	9	8		1	4.02	
		S 9016	P	10	10			3.89	
		S 9017	P	13	13			3.68	
		S 9018	P	6	6			3.57	
		S 9019	P	13	13			3.45	
		S 9020	P	13	12		1	3.72	
		S 9021	NP	0				--	
		S 9022	P	14	14			6.07	
		S 9023	P	12	12			3.80	
		S 9024	NP	0				--	

\* P = Pregnant; NP = Not Pregnant

## FOOD AND DRUG RESEARCH LABORATORIES, INC.

Group 2  
 Material Aspirin  
 Dose 250 mg/kg

Date February 15, 1972  
 Reproduction Data in Rats (Individual)  
 Laboratory No. 0729

## Appendix II

Dam No.	Fate*	Implant Sites	Fetuses Alive	Fetuses Dead	Resorption Sites	Average Fetus Weight (g)	Remarks
A 9001	NP	0				--	Died Day 16.
A 9002	NP	0				--	Died Day 19.
A 9003	P	8				3.52	
A 9004	NP	0				--	Died Day 15.
A 9005	NP	0				--	
A 9006	P	10				3.29	
A 9007	P	11				3.37	
A 9008	P	14			1	3.39	
A 9009	P	10	8		2	3.49	
A 9010	P	14				3.13	
A 9011	P	11				5.13	
A 9012	P	12			1	3.27	
A 9013	P	16			1	3.48	
A 9014	NP	0				--	
A 9015	P	12				3.48	
A 9016	P	11			2	--	
A 9017	P	11				3.70	
A 9018	P	14				3.59	
A 9019	P	13			3	3.77	
A 9020	P	12				3.62	
A 9021	P	13				3.20	
A 9022	NP	0				--	
A 9023	P	10				3.39	
A 9024	P	8				3.41	
A 9025	P	5			1	3.57	
A 9026	P	14			4	3.73	

\* P = Pregnant; NP = Not Pregnant

## FOOD AND DRUG RESEARCH LABORATORIES, INC.

Group 7  
 Material FDA 71-23  
 Dose 27 mg/kg

Date February 15, 1972  
 Reproduction Data in Rats (Individual)  
Laboratory No. 0729 g

## Appendix II

Dam No.	Fate*	Implant Sites	Fetuses		Resorption Sites	Average Fetus Weight (g)	Remarks
			Alive	Dead			
G 9001	P	11	10	1		3.78	
G 9002	P	10	9		1	3.93	
G 9003	P	14	13		1	4.03	
G 9004	P	11	11			3.54	
G 9005	P	9	8		1	3.87	
G 9006	P	11	11			3.87	
G 9007	P	10	10			3.76	
G 9008	P	9	7		2	3.84	
G 9009	P	10	10			4.05	
G 9010	P	11	11			3.83	
G 9011	P	9	9			3.80	
G 9012	P	8	8			3.50	
G 9013	P	14	13		1	3.69	
G 9014	P	14	14			3.78	
G 9015	P	10	9		1	4.17	
G 9016	P	7	4		3	3.61	
G 9017	P	10	10			3.68	
G 9018	P	9	5		4	5.10	
G 9019	P	13	13			3.75	
G 9020	P	10	10			4.22	
G 9021	P	10	10			5.46	
G 9022	P	11	11			3.97	
G 9023	P	11	11			5.57	
G 9024	P	12	10		2	3.29	

\* P = Pregnant; NP = Not Pregnant

## FOOD AND DRUG RESEARCH LABORATORIES, INC.

Group 8  
 Material FDA 71-23  
 Dose 90 mg/kg

Date February 15, 1972  
 Reproduction Data in Rats (Individual)  
Laboratory No. 0729 g

## Appendix II

Dam No.	Fate*	Implant Sites	Fetuses Alive	Fetuses Dead	Resorption Sites	Average Fetus Weight (g)	Remarks
G 9031	P	12	12	—	—	4.23	
G 9032	NP	0	—	—	—	—	
G 9033	P	12	10	2	—	4.03	
G 9034	P	12	12	—	—	3.24	
G 9035	NP	0	—	—	—	—	
G 9036	P	14	12	2	—	4.03	
G 9037	P	9	9	—	—	4.00	
G 9038	P	11	11	—	—	3.77	
G 9039	P	8	6	2	—	3.64	
G 9040	P	9	9	—	—	3.92	
G 9041	NP	0	—	—	—	—	
G 9042	P	12	10	2	—	3.66	
G 9043	P	10	10	—	—	3.67	
G 9044	P	14	13	1	—	3.75	
G 9045	NP	0	—	—	—	—	
G 9046	NP	0	—	—	—	—	
G 9047	P	11	10	1	—	5.23	
G 9048	P	11	11	—	—	3.53	
G 9049	P	11	11	—	—	3.43	
G 9050	P	15	15	—	—	4.05	
G 9051	P	11	11	—	—	3.90	
G 9052	P	10	10	—	—	3.68	
G 9053	P	15	15	—	—	3.83	
G 9054	P	11	9	2	—	3.25	
G 9055	P	10	10	—	—	3.78	
G 9056	P	9	9	—	—	3.98	

\* P = Pregnant; NP = Not Pregnant

## FOOD AND DRUG RESEARCH LABORATORIES, INC.

Group 9  
 Material FDA 71-23  
 Dose 300 mg/kg

Date February 15, 1972  
 Reproduction Data in Rats (Individual)  
 Laboratory No. 0729 8

## Appendix II

Dam No.	Fate*	Implant Sites	Fetuses Alive	Fetuses Dead	Resorption Sites	Average Fetus Weight (g)	Remarks
G 9061	NP	0	15	0	0	4.15	--
G 9062	P	15	0	0	0	--	--
G 9063	NP	0	10	0	0	3.47	
G 9064	P	10	10	0	0	3.99	
G 9065	P	11	11	0	0	4.06	
G 9066	P	15	15	0	0	2.43	
G 9067	P	12	8	0	2	2.43	
G 9068	P	11	11	0	0	3.97	
G 9069	P	3	3	0	0	3.94	
G 9070	P	11	9	0	0	3.85	
G 9071	P	14	13	1	1	3.94	
G 9072	P	12	12	0	0	3.39	
G 9073	P	11	10	1	1	3.86	
G 9074	P	14	13	1	1	4.09	
G 9075	NP	0	0	0	0	--	--
G 9076	P	12	12	0	0	3.95	
G 9077	P	14	14	0	0	3.87	
G 9078	P	12	11	1	1	3.64	
G 9079	P	11	2	0	0	3.79	
G 9080	P	10	8	0	1	3.79	
G 9081	P	11	11	0	0	3.99	
G 9082	P	10	9	0	0	3.87	
G 9083	P	10	10	0	0	3.54	
G 9084	P	11	11	0	0	3.30	
G 9085	P	14	10	4	4	4.00	

\* P = Pregnant; NP = Not Pregnant

## FOOD AND DRUG RESEARCH LABORATORIES, INC.

Group 9 Date February 15, 1972  
 Material FDA 71-23 Laboratory No. 0731 8  
 Dose 300 mg/kg

## Appendix II

Reproduction Data in Rabbits (Individual)

Fate\*      Corpora Lutea

Implant Sites

Fetuses Alive Dead

Resorption Sites

Average Fetus Weight (g)

Remarks

G 1031	P	11	4	3	1	37.1	
G 1032	P	8	7	7		38.8	
G 1033	P	8	7	7		26.6	
G 1034	P	5	5	5		32.7	
G 1035	P	10	5	5		41.4	1 Neonatal death.
G 1036	P	5	1	1		35.4	1 Neonatal death.
G 1037	P	7	7	7		31.6	
G 1038	P	9	8	8		27.1	
G 1039	P	7	7	7		36.8	
G 1040	P	12	6	5	1	36.7	---
G 1041	P	7	6	6		41.7	Aborted Day 28; destroyed pups
G 1042	P	5	4	3		---	
G 1043	P	3	0	0		---	
G 1044	NP	0	0	0		---	Died Day 11.
G 1045	P	6	2	2		45.9	

## FOOD AND DRUG RESEARCH LABORATORIES, INC.

Group 10  
 Material FDA 71-23  
 Dose 1000 mg/kg

## Appendix II

Reproduction Data in Rabbits (Individual)      Date February 15, 1972

Dam No.	Fate*	Corpora Lutea	Implant Sites	Alive	Fetuses Dead	Resorption Sites	Average Fetus Weight (g)	Remarks
G 1046	P	7	7	5	1		36.5	Aborted Day 27.
G 1047	P	11	7	6			---	
G 1048	NP	0	0				---	Aborted Day 26.
G 1049	P	12	3	3			---	
G 1050	NP	4	0				---	
G 1051	P	7	6	6			---	Died Day 10.
G 1052	P	5	2	2			37.8	
G 1053	P	8	7	7			37.6	
G 1054	P	9	6	6			21.3	
G 1055	P	7	6	5		1	38.7	Died Day 13.
G 1056	P	4	4	4			---	
G 1057	P	7	5	5			31.6	
G 1058	NP	0	0				---	
G 1059	P	2	2	2			40.8	
G 1060	P	7	3	2		1	42.4	

\* P = Pregnant;    NP = Not Pregnant

## FOOD AND DRUG RESEARCH LABORATORIES, INC.

Group 8

Appendix II

Date February 15, 1972Material FDA 71-23

Reproduction Data in Rabbits (Individual)

Laboratory No. 0731 gDose 90 mg/kg

Dam No.	Fate*	Corpora Lutea	Implant Sites	<u>Fetuses</u>	Resorption Sites	Average Fetus Weight (g)	Remarks
				<u>Alive</u>	<u>Dead</u>		
G 1016	P	11	9	6		29.3	
G 1017	P	11	7	7		37.8	
G 1018	P	7	4	4		41.3	
G 1019	NP	0	0			---	
G 1020	NP	6	0			---	
G 1021	NP	0	0			---	
G 1022	P	6	3			38.9	
G 1023	P	11	7	5		41.5	
G 1024	P	7	6	4		28.7	
G 1025	P	5	5	4		37.8	
G 1026	NP	0	0			---	
G 1027	P	5	3			39.4	
G 1028	P	6	5			38.9	
G 1029	P	7	6			---	
G 1030	P	5	3			32.3	Aborted Day 26; pups destroyed
				2			

\* P = Pregnant; NP = Not Pregnant

## FOOD AND DRUG RESEARCH LABORATORIES, INC.

Group 7  
 Material FDA 71-23  
 Dose 27 mg/kg

Date February 15, 1972  
 Reproduction Data in Rabbits (Individual) Laboratory No. 0731 g

## Appendix II

Dam No.	Fate*	Corpora Lutea	Implant Sites	Fetuses Alive	Fetuses Dead	Resorption Sites	Average Fetus Weight (g)	Remarks
G 1001	P	10	8	1			42.8	6 Neonatal deaths. Died Day 14.
G 1002	P	1					---	---
G 1003	P	8	4				---	---
G 1004	NP	3	0					
G 1005	P	6	1				47.0	
G 1006	P	6	3				41.7	
G 1007	P	7	6				41.2	
G 1008	NP	12	0					
G 1009	P	10	6				28.9	
G 1010	P	9	4				39.2	
G 1011	NP	2	0					
G 1012	P	4	1				49.5	
G 1013	P	9	2				49.2	
G 1014	NP	0	0					
G 1015	P		5				31.3	1 Neonatal death.

\* P = Pregnant; NP = Not Pregnant

## FOOD AND DRUG RESEARCH LABORATORIES, INC.

Group 2  
 Material 6-Amino nicotinamide  
 Dose 5 mg/kg

Date February 15, 1972  
 Reproduction Data in Rabbits (Individual) Laboratory No. 0731

## Appendix II

Dam No.	Fate*	Corpora Lutea	Implant Sites	Alive	Fetuses Dead	Resorption Sites	Average Fetus Weight (g)	Remarks
Z 1001	P	2	2	2	0	2	---	
Z 1002	P	5	2	2	0	2	---	
Z 1003	P	5	4	4	0	4	---	
Z 1004	P	3	1	1	5	1	24.1	1 Neonatal death.
Z 1005	P	9	1	1	0	3	20.1	2 Neonatal deaths.
Z 1006	NP	4	0	10	2	1	---	1 Neonatal death.
Z 1007	P	10	2	1	0	4	4	Died day 22.
Z 1008	P	6	4	6	0	0	---	
Z 1009	NP	5	0	5	0	0	---	
Z 1010	NP	4	0	4	0	0	4	
Z 1011	P	4	0	0	0	4	38.6	
Z 1012	P	12	4	12	4	2	37.3	
Z 1013	P	10	6	10	6	0	---	
Z 1014	NP	5	3	5	0	0	---	
Z 1015	NP	3	0	3	0	0	---	

## FOOD AND DRUG RESEARCH LABORATORIES, INC.

Group 1  
Material Sham  
Dose 0

Date February 15, 1972  
Appendix II  
Reproduction Data in Rabbits (Individual) Laboratory No. 0731

Dam No.	Fate*	Corpora Lutea	Implant Sites	Alive	Dead	Fetuses	Resorption Sites	Average Fetus Weight (g)	Remarks
S 1001	P	13	3	3	0	1	1	38.4	
S 1002	P	6	2	1	1			36.1	
S 1003	NP	0	0					---	
S 1004	NP	4	0					---	
S 1005	P	9	6	5	1			40.3	
S 1006	P	13	3	2	1			43.9	
S 1007	P	7	7	7	0			---	
S 1008	NP	0	0	4	4			---	
S 1009	P	5	4	4	0			---	
S 1010	NP	0	0	0				---	
S 1011	P	4	2	1	1	2	2	40.7	
S 1012	P	17	5	2	3			43.7	
S 1013	P	13	4	2	1	2	2	---	
S 1014	P	5	3	3	0	3	3	---	
S 1015	NP	10	0					---	

\* P = Pregnant; NP = Not Pregnant;



each litter were then placed in an incubator for 24 hours for the evaluation of neonatal survival. All surviving pups were sacrificed, and all pups examined for visceral abnormalities (by dissection). All fetuses were then cleared in potassium hydroxide (KOH), stained with alizarin red S dye and examined for skeletal defects.



## Appendix I

### Teratology Study in Rabbits

Virgin, adult, Dutch-belted female rabbits were individually housed in mesh bottom cages in temperature and humidity-controlled quarters with free access to food and fresh tap water. On Day 0, each doe was given an injection of 0.4 ml of human chorionic gonadotropin (400 IU) via the marginal ear vein. Three hours later, each doe was inseminated artificially with 0.3 ml of diluted semen from a proven donor buck using approximately  $20 \times 10^6$  motile sperm according to the procedure described by Vogen et al (Pharmacologist 11, 282 (1969)). Beginning on Day 6 and continuing daily through Day 18 the females were dosed with the indicated dosages by oral intubation; the controls were sham treated.

Body weights were recorded on Days 0, 6, 12, 18, and 29 of gestation. All animals were observed daily for appearance and behavior, with particular attention to food consumption and body weight in order to rule out any abnormalities which may have occurred as a result of anorexic effects in the pregnant female animal.

On Day 29 all does were subjected to Caesarean section under surgical anesthesia, and the numbers of corpora lutea, implantation sites, resorption sites and live and dead fetuses were recorded. Body weights of the live pups were also recorded. The urogenital tract of each animal was examined in detail for normality. In addition all fetuses underwent a detailed gross examination for the presence of external congenital abnormalities. The live fetuses of

## FOOD AND DRUG RESEARCH LABORATORIES, INC.

Groups 1 &amp; 2; 7 through 10

Species Rabbits

Table 4

Average Body Weights \*

Date February 15, 1972  
Laboratory No. 0731 q

Group	Material	Dose Level	Day -			Day -
			0	6	12	
		mg/kg	kg			
1	Sham	0	2.39	2.36	2.31	2.31 ( 8 )
2	6-AN***	5	2.34	2.43	2.27	2.09 ( 9 )
7	FDA 71-23	27	2.18	2.20	2.24	2.23
8	FDA 71-23	90	2.32	2.34	2.33	2.34
9	FDA 71-23	300	2.26	2.29	2.30	2.30
10	FDA 71-23	1000	2.43	2.46	2.38	2.45 ( 8 )

\* Of pregnant dams.

\*\* Number of surviving dams in parentheses (c.f. Table 1).

\*\*\* 6-amino nicotinamide dosed on Day 9.

FOOD and DRUG RESEARCH LABORATORIES, INC.

Groups 1 & 2, 7 through 10

Material FDA 71-23

Table 3

Summary of Skeletal Findings  
(Rabbits)

Laboratory No. 0731 q  
Date February 15, 1972

Findings	Group No:	1	2	7	8	9	10
Live Fetuses Examined	Dose (mg/kg): Sham	6-AN**		27	90	300	1000
Fetuses with: Incomplete sternebrae	7/3	4/3 2/2 2/2	8/5	12/6 4/4	28/9 5/5	11/5 4/3	
Scrambled sternebrae		1/1					
Bipartite sternebrae		2/2					
Missing sternebrae		1/2					
Fused sternebrae		4/4		1/1	2/2	5/4	
Extra sternebrae		1/1					
Fetuses with: Scrambled vertebrae Tail defects (short, scrambled, etc.)		1/1 3/3				1/1	
Fetuses with: Fused ribs Incomplete ribs Wavy ribs Less than 12		2/2 2/2		7/4	5/3	10/7	7/5
Other Findings: Scoliosis Delayed cranial ossification Craniostosis Cleft Palate Anophthalmia		6/3	4/2 2/2 1/1	7/5	11/7	14/9	4/1 7/3

\* Numerator = Number of fetuses affected; Denominator = number of litters affected

Group: 1 & 2: 7 through 10

Food AND DRUG RESEARCH LABORATORIES, INC.

Date February 15, 1.2

Material: FDA 71-23

Table 2  
Reproduction Data  
(Rabbits)

Group:	1	2	7	8	9	10
Dose (mg/kg):	Sham	6-AN*	27	90	300	1000
Number of females:						
Total pregnant	10	10	11	10	14	12
Pregnant at term	8	4	9	10	12	8
Number of corpora lutea:						
Total	106	87	90	89	103	90
Average/pregnant dam	10.6	8.7	8.3	8.9	7.3	7.5
Number of live litters						
Total	6	4	9	10	12	8
Number of implant sites:						
Total	39	30	42	60	72	58
Average/pregnant dam	3.9	3.0	3.8	6.0	6.0	4.8
Number of fetuses alive:						
Total	14	8	36	50	66	49
Average/pregnant dam	1.4	0.80	3.3	5.0	4.7	4.1
Average/live litter	2.33	2.0	4.0	5.0	5.5	6.1
Average/pregnancy at term	1.75	2.0	4.0	5.0	5.5	6.1
Number of fetuses dead:						
Total	12	1	0	0	0	4
Litters with one or more dead	3	1	-	-	-	1
Pregnancies at term (%)	37.5	25.0	-	-	-	12.5
Litters with all dead	2	0	-	-	-	1
Pregnancies at term (%)	25.0	-	-	-	-	12.5
Number of resorptions:						
Total:	13	25	6	10	5	3
Litters with one or more resorptions	7	9	3	5	4	3
Pregnancies at term (%)	87.5	>100	33.3	1.0	33.3	25.0
Litters with total resorptions	2	6	1	0	0	0
Pregnancies at term (%)	25.0	>100	11.0	-	-	-
Average fetus weight, g.	40.5	30.0	41.2	36.6	35.9	31.2

\* Positive control: 5.0 mg/kg 6 - amino nicotinamide dosed on Day 9.

## FOOD AND DRUG RESEARCH LABORATORIES, INC.

Groups: 1 & 2; 7 through 10Material: FDA 71-23

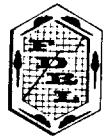
Table 1

Fate Summary  
(Rabbits)Date February 15, 1972  
Laboratory No. 07318

Group	Material	Dose mg/kg	Mated	Total	Pregnant	At Term	
						Surviving (Total)	Number Pregnant
1	Sham	0		15		10	13
2	6-AN*	5		15		10	14
7	FDA 71-23	27		15		11	14
8	FDA 71-23	90		15		11	14
9	FDA 71-23	300		15		14	13
10	FDA 71-23	1000		15		12	11
							8

\* Positive Control: 6 - Amino nicotinamide dosed on Day 9.

**Food and Drug Research Laboratories**  
INCORPORATED



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Telephone: TWining 4-0800  
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FINAL  
REPORT

Submitted to: DHEW/Public Health Service  
Food and Drug Administration CA-272  
5600 Fishers Lane-Room 5C-13  
Rockville, Maryland 20852

Date: February 15, 1972  
Laboratory No. 0731 g  
Contract No. FDA 71-260

Sample: Fine dark purple material.

Marking: FDA 71-23 (Amaranth; FD & C Red No. 2)

Examination Requested: Teratologic evaluation of FDA 71- 23 in rabbits

Procedure: (See Appendix I)

Results: See Tables 1 through 4 and Appendix II

Conclusion: Attention is called to the fact that this is the first of a series of reports which will be issued in accordance with the terms of the contract cited above. Eventually, a total of at least 36 compounds will have been tested in 18 pairs; each pair being run concurrently against one sham-treated control and one positive control group. Because of the inherent variability of biological data of the type dealt with here, the accumulation and pooling of sequential sets of control values will greatly enhance the statistical value of the findings and the ultimate reliability of the test results.

For these reasons, the conclusion stated below is regarded as provisional and subject to reexamination in the light of later findings:

"The administration of up to 1000 mg/kg (body weight) of the test material to pregnant rabbits for 13 consecutive days had no clearly discernible effect on nidation or on maternal or fetal survival. The number of abnormalities seen in either soft or skeletal tissues of the test groups did not differ from the number occurring spontaneously in the sham-treated controls."

FOOD AND DRUG RESEARCH LABORATORIES, INC.

*Kenneth Morgareidge*  
Kenneth Morgareidge, Ph.D.  
Vice President

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## FOOD AND DRUG RESEARCH LABORATORIES, INC.

Group 10      Material FDA 71-23      Dose 1000 mg/kg

Date February 15, 1972      Reproduction Data in Hamsters (Individual)

Laboratory No. 0730 g

Dam No.	Fate*	Implant Sites	Fetuses		Resorption Sites	Average Fetus Weight (g)	Remarks
			Alive	Dead			
G 0091	P	12	12			2.08	
G 0092	P	13	13			1.67	
G 0093	P	8	7		1	1.75	
G 0094	P	15	15			1.61	
G 0095	P	12	12			1.74	
G 0096	NP	0				--	
G 0097	P	17	17			1.84	
G 0098	P	10	7			1.95	
G 0099	P	13	13			1.85	
G 0100	P	11	11			1.32	
G 0101	P	14	13			1.80	
G 0102	P	16	13			1.84	
G 0103	P	12	11			1.75	
G 0104	P	16	16			1.87	
G 0105	P	13	13			1.70	
G 0106	--	--	--			--	
G 0107	P	14	14			2.00	
G 0108	NP	0				--	
G 0109	P	12	10			1.21	
G 0110	P	13	13			1.81	
G 0111	P	13	13			1.58	
G 0112	P	13	12			1.64	
G 0113	P	13	12			1.74	

\* P = Pregnant; NP = Not Pregnant

Number not assigned.

## FOOD AND DRUG RESEARCH LABORATORIES, INC.

Group 9  
 Material FDA 71-23  
 Dose 300 mg/kg

Date February 15, 1972  
 Laboratory No. 0730 g

## Appendix II

## Reproduction Data in Hamsters (Individual)

Dam No.	Fate *	Implant Sites	Fetuses Alive Dead	Resorption Sites	Average Fetus Weight (g)	Remarks
G 0061	P	11	11		1.87	
G 0062	P	11	11		2.07	
G 0063	P	9	8	1	1.82	
G 0064	P	14	14		1.91	
G 0065	P	14	14		1.58	
G 0066	P	13	13		1.70	
G 0067	P	16	16		2.09	
G 0068	P	12	12		1.87	
G 0069	P	15	12	2	1.14	
G 0070	P	12	12	1	1.86	
G 0071	P	15	15		2.00	
G 0072	P	14	12	2	1.96	
G 0073	P	15	15		1.74	
G 0074	P	13	12		1.70	
G 0075	P	11	11	1	1.83	
G 0076	P	13	11	13	--	
G 0077	P	11	11	1	1.83	
G 0078	P	14	13		1.82	
G 0079	P	12	11	1	1.47	
G 0080	NP	0	10	2	1.91	
G 0081	P	12	10		--	
G 0082	NP	0	10		1.87	
G 0083	P	10	10		2.13	
G 0084	P	15				

\* P = Pregnant; NP = Not Pregnant

## FOOD AND DRUG RESEARCH LABORATORIES, INC.

Group 8  
 Material FDA 71-23  
 Dose 90 mg/kg

Date February 15, 1972  
 Reproduction Data in Hamsters (Individual) Laboratory No. 0730 g

## Appendix II

Dam No.	Fate*	Implant Sites	Fetuses Alive	Fetuses Dead	Resorption Sites	Average Fetus Weight (g)	Remarks
G 0031	P	11	11			1.81	
G 0032	P	17	17			1.76	
G 0033	P	14	13	1	1	1.84	
G 0034	P	13	13			1.82	
G 0035	P	12	11	1	1	1.78	
G 0036	P	12	11	1	1	2.09	
G 0037	P	15	15			1.85	
G 0038	P	13	12	1	1	1.88	
G 0039	P	11	11			1.08	
G 0040	P	14	13	1	1	1.76	
G 0041	P	11	11			2.02	
G 0042	P	15	15			1.78	
G 0043	P	14	14			1.87	
G 0044	P	15	14	1	1	1.68	
G 0045	P	12	11	1	1	1.69	
G 0046	P	13	13			1.63	
G 0047	P	11	11			1.84	
G 0048	P	11	11			2.06	
G 0049	P	13	13			1.63	
G 0050	P	13	12	1	1	1.69	
G 0051	P	10	10	10	1	--	
G 0052	P	14	14	4	4	1.94	
G 0053	P	14	10	4	4	1.80	
G 0054	P	13	13	4	4	1.85	

\* P = Pregnant; NP = Not Pregnant

## FOOD AND DRUG RESEARCH LABORATORIES, INC.

Group 7      Material FDA 71-23      Dose 27 mg/kg

Appendix II      Reproduction Data in Hamsters (Individual)

Date February 15, 1972      Laboratory No. 0730 g

Dam No.	Fate*	Implant Sites	Fetuses Alive	Fetuses Dead	Resorption Sites	Average Fetus Weight (g)	Remarks
G 0001	P	14	13			1.52	
G 0002	P	9	9			2.37	
G 0003	P	11	11			1.83	
G 0004	P	13	13			1.70	
G 0005	P	12	12			1.69	
G 0006	P	10	10			1.69	
G 0007	P	11	11			1.97	
G 0008	P	13	12			2.02	
G 0009	P	13	10			1.16	
G 0010	P	12	12			1.56	
G 0011	P	14	13			1.86	
G 0012	P	13	13			1.89	
G 0013	P	14	14			1.40	
G 0014	P	13	13			1.74	
G 0015	P	11	10			1.55	
G 0016	P	13	7			1.77	
G 0017	P	12	11			2.05	
G 0018	P	14	14			2.15	
G 0019	P	11	10			1.59	
G 0020	P	10	10			1.73	
G 0021	P	11	11			2.03	
G 0022	P	10	8			2.09	
G 0023	P	13	13			1.84	
G 0024	NP	0				--	

\* P = Pregnant; NP = Not Pregnant

## FOOD AND DRUG RESEARCH LABORATORIES, INC.

Group 2      Date February 15, 1972  
 Material Aspirin      Reproduction Data in Hamsters (Individual)  
 Dose 250 mg/kg      Laboratory No. 0730

## Appendix II

Dam No.	Fate*	Implant Sites	Fetuses Alive	Fetuses Dead	Resorption Sites	Average Fetus Weight (g)	Remarks
A 0001	P	13				1.75	
A 0002	NP	0				--	
A 0003	P	13				1.82	
A 0004	P	12	11	1		2.12	
A 0005	P	12	12			1.90	
A 0006	NP	0				--	
A 0007	P	15	15			2.10	
A 0008	NP	0				--	
A 0009	P	13				1.67	
A 0010	P	12	12			1.04	
A 0011	P	14	14			1.91	
A 0012	P	11	11			1.67	
A 0013	P	11	10	1	1	1.94	
A 0014	P	14	13	1	1	1.82	
A 0015	P	16	16	1	1	1.80	
A 0016	P	12	12	1	1	1.61	
A 0017	P	13	13	1	1	1.89	
A 0018	P	13	13	1	1	1.87	
A 0019	P	12	12	1	1	1.74	
A 0020	P	12	12	1	1	1.84	
A 0021	P	14	14	1	1	1.62	
A 0022	P	11	11	1	1	1.69	
A 0023	P	10	8	2	2	1.86	
A 0024	P	9	9	2	2	2.42	

\* P = Pregnant; NP = Not Pregnant

## FOOD AND DRUG RESEARCH LABORATORIES, INC.

Group 1 Date February 15, 1972  
 Material Sham Reproduction Data in Hamsters (Individual) Laboratory No. 0730

## Appendix II

Dose 0

Dam No.	Fate*	Implant Sites	Fetuses Alive	Fetuses Dead	Resorption Sites	Average Fetus Weight (g)	Remarks
S 0001	P	12	12			1.62	
S 0002	P	12	12			1.82	
S 0003	P	9	9			1.66	
S 0004	P	14	14			2.01	
S 0005	P	17	15			1.97	
S 0006	P	14	14			2.06	
S 0007	P	10	10			1.94	
S 0008	P	14	12		2	1.85	
S 0009	P	12	12			1.03	
S 0010	P	13	13			1.78	
S 0011	P	16	15		1	1.63	
S 0012	P	14	13		1	1.88	
S 0013	P	14	14			1.80	
S 0014	P	17	14			2.00	
S 0015	P	17	17			0.98	
S 0016	P	10	10			1.70	
S 0017	P	15	15			1.65	
S 0018	P	10	10			1.84	
S 0019	P	16	16			1.68	
S 0020	P	12	12			--	
S 0021	NP	0	0			1.77	
S 0022	P	10	9			1.55	
S 0023	P	9	9			--	
S 0024	P	11	11				

\* P = Pregnant; NP = Not Pregnant



## Appendix I

### Teratology Study in Hamsters

Virgin adult female golden hamsters from an outbred strain were individually housed in mesh bottom cages in temperature and humidity controlled quarters with free access to food and fresh tap water at all times. They were mated (1 to 1) with mature males and the appearance of motile sperm in the vaginal smear was considered as Day 0 of gestation. Beginning on Day 6 and continuing daily through Day 10 of gestation, the indicated dose levels of the test material were administered by oral intubation; the controls were sham-treated.

Body weights were recorded on Days 0, 8, 10, and 14 of the gestation period. All animals were observed daily for appearance and behavior with particular attention to food consumption in order to better recognize any abnormalities resulting from anorexic effects in the pregnant animal.

On Day 15, all animals were subjected to Caesarian section under deep anesthesia and the numbers of implantation sites, resorption sites, live and dead fetuses were recorded. All live pups were weighed and the genital tract of each dam was examined for any anatomical abnormalities.

All fetuses were examined grossly for the presence of external congenital defects and one-third of each litter underwent detailed visceral examination under 10X magnification. The remaining two-thirds of the pups were cleared in potassium hydroxide, stained with alizarin red dye, and examined for the presence of skeletal abnormalities.

## FOOD AND DRUG RESEARCH LABORATORIES, INC.

Groups 1 & 2; 7 through 10Species HamstersDate February 15, 1972

Table 4

Average Body Weights\*Laboratory No. 0730 g

Group	Material	Dose Level	mg/kg	Day				14**
				0	6	8	10	
1	Sham	0	106.2	110.2	113.7	124.7	144.8	(21)
2	Aspirin	250	104.9	109.3	112.6	121.8	144.8	(21)
7	FDA 71-23	27	110.6	113.1	116.6	129.1	146.3	(23)
8	FDA 71-23	90	109.5	113.6	117.8	128.9	149.1	(24)
9	FDA 71-23	300	110.3	115.1	120.1	130.6	151.5	(22)
10	FDA 71-23	1000	110.3	115.3	119.2	132.0	152.5	(20)

\* of pregnant dams

\*\* Number of surviving dams in parentheses (c.f. Table 1)

Groups 1&2; 7 through 10  
Material FDA 71-23

FOOD and DRUG RESEARCH LABORATORIES, INC.

Laboratory No. 0730 g  
Date February 15, 1972

Table 3

Summary of Skeletal Findings  
(Hamsters)

Findings	Group No:	1	2	7	8	9	10
	Dose (mg/kg):Sham	Aspirin		90	300	1000	
<u>Live Fetuses Examined</u>		189/21	184/21	188/23	204/23	187/21	169/19
<u>Fetuses with:</u>							
Incomplete sternebrae	17/6	20/10	42/11	29/13	37/12		32/11
Scrambled sternebrae	115/19	91/19	112/22	108/19	102/19		106/18
Bipartite sternebrae							
Missing sternebrae							
Fused sternebrae							
Extra sternebrae							
Incomplete hyoid							
<u>Fetuses with:</u>							
Scrambled vertebrae							
Tail defects (short, scrambled, etc.)	54/12	82/17	123/18				
Incomplete ossification; vertebrae				1/1			
Extra vertebrae				1/1			
Club foot				1/1			
<u>Fetuses with:</u>							
Fused ribs	1/1	1/1	2/2				
Incomplete ribs	5/4		6/3				
Wavy ribs							
Less than 12 ribs				1/1			
More than 13 ribs				16/9	18/10		
Anophthalmia					2/2		
<u>Other Findings:</u>							
Scoliosis	17/2	10/2	45/11	28/10	38/10		61/12
Delayed cranial ossification ~				1/1			
Craniosclerosis				1/1			
Feeble; retarded ossification	9/2			20/7			10/3
Meningocele				2/2			

\* Numerator = Number of fetuses affected; Denominator = number of litters affected

Group: 1 & 2; 7 through 10

Material: FDA 71-23

FOOD AND DRUG RESEARCH LABORATORIES, INC.

Date February 15, 1972

Table 2

Reproduction Data  
( Hamsters )

Group:	1	2	Aspirin*	7	8	9	10
Dose (mg/kg):	Sham			27	90	300	1000
Number of females:							
Total pregnant	22	21		23	24	22	20
Pregnant at term	21	21		23	23	21	20
Number of live litters:							
	21	21		23	23	21	20
Number of implant sites:							
Total	281	262		277	311	282	260
Average/pregnant dam	12.8	12.5		12.0	13.0	12.8	13.0
Number of fetuses alive:							
Total	264	257		260	289	258	247
Average/live litter	12.6	12.2		11.3	12.6	12.3	12.4
Average/pregnancy at term	12.6	12.2		11.3	12.6	12.3	12.4
Number of fetuses dead:							
Total	0	1		2	0	3	2
Litters with one or more dead	0	1		2	0	2	2
Pregnancies at term (%)	0	4.8		8.7	0	9.5	10.0
Litters with all dead	0	0		0	0	0	0
Pregnancies at term (%)	0	0		0	0	0	0
Number of resorptions:							
Total:	17	4		15	22	21	11
Litters with one or more resorptions	5	3		8	10	7	7
Pregnancies at term (%)	23.8	14.3		34.8	43.5	33.3	35.0
Litters with total resorptions	1	0		0	1	1	0
Pregnancies at term (%)	4.8	0		0	4.3	4.8	0
Average fetus weight, g.	1.72	1.81		1.79	1.79	1.82	1.74

\* Positive control at 250 mg/kg

## FOOD AND DRUG RESEARCH LABORATORIES, INC.

Groups: 1 & 2; 7 through 10Material: FDA 71-23

Table 1

Fate Summary  
(Hamsters)Date February 15, 1972  
Laboratory No. 0730 8

Group	Material	Dose mg/kg	Mated	Total	At Term	
					Pregnant	Surviving (Total) Number Pregnant
1	Sham	0.	23	22	23	21
2	Aspirin*	250	24	21	24	21
7	FDA 71-23	27	24	23	24	23
8	FDA 71-23	90	24	24	24	23
9	FDA 71-23	300	24	22	24	21
10	FDA 71-23	1000	22	20	22	20

\* Positive Control

**Food and Drug Research Laboratories**  
INCORPORATED



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Telephone: TWining 4-0800  
Cable: Foodlabs, New York

**FINAL  
REPORT**

Submitted to: DHEW/Public Health Service  
Food and Drug Administration CA-272  
5600 Fishers Lane-Room 5C-13  
Rockville, Maryland 20852

Date: February 15, 1972

Laboratory No. 0730 g  
Contract No. FDA 71-260

Sample: Fine dark purple material.

Marking: FDA 71-23 (Amaranth: FD & C Red No. 2).

Examination Requested: Teratologic evaluation of FDA 71- 23 in hamsters

Procedure: See Appendix I

Results: See Tables 1 through 4 and Appendix II

Conclusion: Attention is called to the fact that this is the first of a series of reports which will be issued in accordance with the terms of the contract cited above. Eventually, a total of at least 36 compounds will have been tested in 18 pairs; each pair being run concurrently against one sham-treated control and one positive control group. Because of the inherent variability of biological data of the type dealt with here, the accumulation and pooling of sequential sets of control values will greatly enhance the statistical value of the findings and the ultimate reliability of the test results.

For these reasons, the conclusion stated below is regarded as provisional and subject to reexamination in the light of later findings:

"The administration of up to 1000 mg/kg (body weight) of the test material to pregnant hamsters for 5 consecutive days had no clearly discernible effect on nidation or on maternal or fetal survival. The number of abnormalities seen in either soft or skeletal tissues of the test groups did not differ from the number occurring spontaneously in the sham-treated controls."

FOOD AND DRUG RESEARCH LABORATORIES, INC.

*Kenneth Morganridge*  
Kenneth Morganridge, Ph.D.  
Vice President

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## FOOD AND DRUG RESEARCH LABORATORIES, INC.

Group 10  
 Material FDA 71-23  
 Dose 1000 mg/kg

## Appendix II

Date February 15, 1972  
 Laboratory No. 0729 8

## Reproduction Data in Rats (Individual)

Dam No.	Fate*	Implant Sites	Fetuses	Resorption Sites	Average Fetus Weight (g)	Remarks
			Alive Dead			
G 9091	P	13	13		3.48	
G 9092	P	12	10		3.69	
G 9093	P	11	11		3.98	
G 9094	P	8	8		3.55	
G 9095	NP	0			--	
G 9096	NP	0			--	
G 9097	P	10	10		3.81	
G 9098	P	13	13		3.39	
G 9099	P	10	9		3.91	
G 9100	P	12	12		4.01	
G 9101	P	15	14	1	4.03	
G 9102	P	8	8		3.56	
G 9103	P	12	10		3.97	
G 9104	P	10	9		3.92	
G 9105	P	4	3		3.91	
G 9106	P	1	1		4.65	
G 9107	P	10	8	2	3.78	
G 9108	P	12	12		3.51	
G 9109	NP	0			--	
G 9110	P	9	8		4.30	
G 9111	P	10	10		--	
G 9112	P	11	11		3.81	
G 9113	P	13	13		3.92	
G 9114	P	13	12		3.43	
G 9115	P	10	8		3.72	

\* P = Pregnant; NP = Not Pregnant